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METHANE AND CARBON DIOXIDE FLUXES FROM *LIMONIUM* RESIDUES DECOMPOSITION IN SALTMARSH SOILS: EFFECTS OF TIDE REGIME

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Abstract

The flooding regime of saltmarshes strongly affects organic matter mineralisation, representing a unique situation where oxygen diffusion is either impeded by submersion or favoured by retreating water in regular cycles within the same day. Decomposition of *Limonium vulgare* Mill. residues in saltmarsh soils was evaluated measuring CO_2 and CH_4 emissions. Four different saltmarshes from the Grado Lagoon (Northern Adriatic Sea) were investigated. Soils were characterised by a similar vegetation (*Sarcocornietea* class) and similar high coverage of *L. vulgare* (70-75%) but differed in redox potential, texture and organic carbon content. Hydromorphic conditions were reproduced in mesocosms, and soils were incubated under fully aerobic, fully anaerobic and transient (6 hours cycles) tidal states. Partially decomposed litter (leaves) of *L. vulgare* was added and decomposition processes were monitored through CO_2 and CH_4 emissions. Larger CO_2 emissions were measured under aerobic conditions, in particular in soil samples with coarse texture. Fully anoxic and tidal regimes showed a similar behaviour. On the contrary, CH_4 emissions were less dependent upon flooding, showing only slightly larger values under completely submerged conditions. Larger CH_4 emissions have been obtained in fine textured soils. Soil organic matter content also influenced gas emissions: larger values corresponded to higher emissions of both CO_2 and CH_4.

Key words: *saltmarsh soils, hydromorphic conditions, mineralisation, methane.*

Introduction

Wetlands, especially saltmarshes, hold a key role in the global carbon (C) cycle, being both a potential sink and a source of C in the exchange among terrestrial and aquatic ecosystems and the atmosphere (Whiting and Chanton, 2001). Erosion processes and the raising of the medium sea level could increase the C losses from saltmarshes (Voss et al., 2013; DeLaune and White, 2012). Generally, coastal ecosystems are especially involved in C sequestration processes, due to the high primary production and the low decomposition rates (Valiela et al., 1985). Microorganisms use organic matter as substrate to generate energy and release C mainly in the form of CO_2 (Willd, 1988). Decomposition processes are controlled by many factors among which temperature, pH and humidity (Parr and Papendick, 1978), and especially residues and oxygen availability (Kristensen et al., 1995).

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Also the typical alternate flooding regime of saltmarshes strongly affects organic matter decomposition. The slower oxygen diffusion in particular, and the consequent establishment of anoxic conditions in lower areas (Shin et al., 2000), involves a limitation of CO_2 emissions and an increase in CH_4 production by methanogens (Gaillard et al., 1992; Oremland et al., 1982). In fact, saltmarsh soils are saturated by water, in the larger part of the profile. Water acts as a physical barrier for oxygen, that is available in sufficient amounts only in superficial soil layers (Cartaxana and Lloyd, 1999). Anaerobic processes, being lengthy and incomplete, determine a lower release of C in the atmosphere and an increase of C stocked in sediments (Shin et al., 2000). On the other side, CH_4 emissions, in these environments, can be limited by sulfur-reducing bacteria, more efficient in using CO_2 compared to methanogens, as well as by methanotrops when the redox state turns to aerobic conditions (e.g. in superficial soil layers) (Oremland et al., 1982).
Information about specific effects on organic matter mineralisation, of the regular exposure to flooding cycles, is still lacking. The aim of this work was to evaluate decomposition processes of *Limonium vulgare* Mill. residues in saltmarsh soils under three different imposed conditions: fully aerobic, fully anaerobic and simulated tidal conditions. Evaluations were conducted considering CO_2 and CH_4 emissions.

Materials and methods

Study area

For this study, four saltmarshes were selected in the Grado Lagoon (Northern Adriatic sea), at different distances from the open sea: two are located in the inner part of the lagoon, Isole della Gran Chiusa (GC) and Belvedere (BB), one in the outer part, Marina di Macia (MM), and the last in the peri-lagoonal area of recent formation, Banco d'Orio (BO) (Fig. 1).

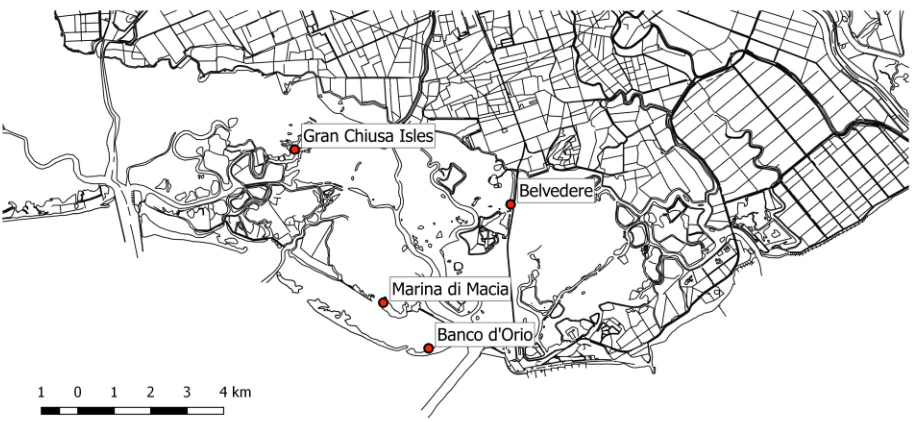

Figure 1. *Saltmarshes selected in the Grado Lagoon*

Sampling sites are characterised by different texture and redox conditions, but similar vegetation (*Sarcocornietea* class) and similar high coverage of *Limonium vulgare* (70-75%). Redox potential (Eh) was measured in the field at each sampling site.

Sampling and soil analysis

Soil cores were collected from each site during spring time, transported to the laboratory and sliced to separate the different horizons. Soil was handled under N_2 to avoid exposure to oxygen. An aliquot was used for analysis of organic C total N, texture and microbial biomass C, and the rest was used for the mesocosms experiment.

Organic C (C_{org}) and total N were analysed with a CHNS Vario MICRO cube. An ASTM 152H Bouyoucos hydrometer was used for texture (Bouyoucos, 1962). Soil microbial biomass carbon was determined using the chloroform fumigation extraction method (Vance et al., 1987) and measured by the automated elemental analyser TOC-V CPN + TNM-1 Analyzer (Shimadzu TOC-VCPN).

Mesocosms

Cylindrical core samples (6 cm d, 5 cm long), reproducing soil profiles, were reconstructed for the mesocosms experiment. Samples were pre-incubated in the dark at 25°C for 4 days. Partially decomposed leaves of *L. vulgare* (5% soil d.w.) were added and soils were further incubated for 12 days. Synthetic marine water was prepared following Kester et al. (1967). Three different flooding conditions were imposed: totally emerged, totally submerged and a 6 hours tidal cycles variation. Hydraulic pumps were used for simulating tides: the water level changed, interchanging emergence and submergence every 6h. After 1, 2, 5, 8 and 13 days from the addition of the residues, 5 ml of the gas phase were collected with a syringe from the sealed mesocosms and stored in screw cap vials equipped with septa. Aliquots (200 µl) of the gas phase were analysed by gas chromatography. An HRGC MEGA 2 series 8540, CE Instruments equipped with a TCD was used for determining CO_2, while a HRGC MEGA 2 capillary column gas chromatograph series 8560, Fisons Instruments, equipped with a FID was used to analyze CH_4.

<u>Results and discussion</u>

Sites characterization

Initial soil Eh values, measured in the field at sampling, differed strongly among sites (Tab. 1): the GC soil recorded the lowest and negative values, in opposition to the BO soil which displayed the highest positive Eh. In BO, the more oxic redox conditions probably depend on the coarser sandy texture, which allows a rapid re-establishment of oxic conditions in this soil during low tide.

In the GC soil, C_{org} as well as microbial biomass and labile C contents were the largest, compared to the other saltmarsh soils.

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Table 1- *Characterization of the four saltmarsh sites considered*

Soils	Eh (mV)	Sand (%)	Silt (%)	Clay (%)	C org (%)	Microbial biomass (μg/g)	Biomass: C org ratio (μg/g)	Labile C (μg/g)
Gran Chiusa Isles (GC)	194 ± 27	52	24	24	6.74 ± 0.60	318	4.72	83.15
Belvedere (BB)	90 ± 47	52	20	28	1.92 ± 0.09	293	15.35	71.57
Marina di Macia (MM)	117 ± 49	72	8	20	2.17 ± 0.12	131	6.08	51.06
Banco d'Orio (BO)	163 ± 21	96	2	2	0.22 ± 0.15	138	64.33	23.55

Larger microbial biomass/C_{org} ratios in BB and BO soils suggest a more recent C accumulation. The BO saltmarsh in particular has a very recent formation compared to the other considered, being located in the peri-lagoonal area of the Grado lagoon, which originated only recently from marine deposits (Fontolan et al., 2012). The BB saltmarsh, originated from dredged materials, has a recent formation too and is about 100 years old.

Mesocosms experiment

Both CH_4 and CO_2 emissions differed greatly among the different soils under tidal variation conditions (Fig. 2).

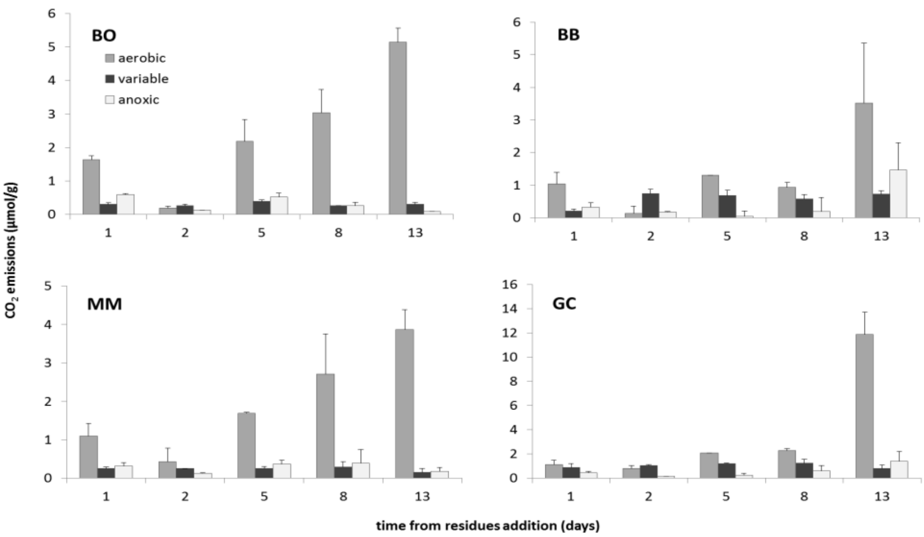

Figure 2. *Cumulated CO_2 emissions during mesocosms incubation under the three different conditions imposed after plant residues addition.*

The large C_{org} content of GC site influenced strongly both CH_4 and CO_2 production: the concentrations in the gas phase were the highest among the four saltmarsh soils throughout the experiment. As expected much larger emissions were obtained from soils just after addition of *Limonium vulgare* residues, due to the large increase of easily decomposable C provided. Comparing the three treatments imposed, significantly larger CO_2 emissions were recorded for amended soils under aerobic conditions, while emissions under transient tidal conditions appeared more similar to those from fully anoxic soils (Fig. 2). This result confirms that the soils situated at medium height in saltmarsh areas, which are affected by regular intermittent flooding, provide a biological environment very close to that of permanently submerged soils.

In coarse textured samples (i.e. MM and BO), CO_2 emissions peaked within less time compared to silty clay soils (i.e. BB and GC). However, the highest values were actually recorded for the GC soil, in which emissions after 13 days of the aerobic incubation were about 3 times larger compared to the other three saltmarsh soils (Fig. 2). This is probably due again to the larger C_{org} and microbial biomass C content measured for this soil (Tab. 1).
CH_4 emissions instead, were relatively similar among treatments and only slightly higher for the anoxic condition (Fig. 3). Similarly to CO_2, texture and C_{org} content exerted an important role also in determining CH_4 emissions, which were larger in fine textured soils rich in organic matter (e.g. GC site), where anoxic conditions were favoured.

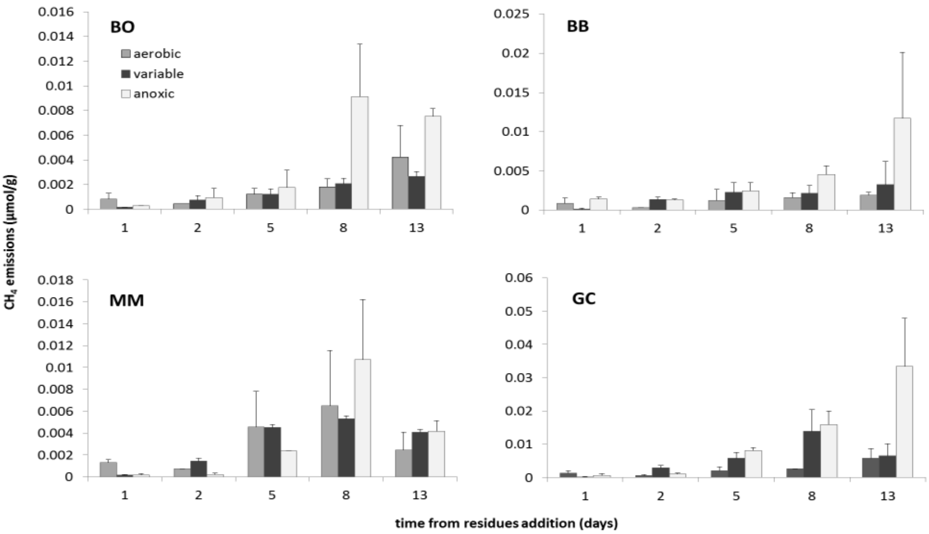

Figure 3. *Cumulated CH_4 emissions during mesocosms incubation under the three different conditions imposed after plant residues addition*

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Eh values were consistently modified during incubation by the addition of residues, clearly distinguishing soils incubated in mesocosms under different regimes, confirming the efficacy of treatments (Fig. 4). Peaks of CO_2 emissions, were much larger in soils which displayed final positive Eh values, while a negative trend was shown for CH_4. A much broader dispersion is observed at positive Eh values and CO_2 peak emissions are much larger in fully aerobic soils, while mineralisation of plant residues in soils subjected to continuous or intermittent periods of submergence appeared more similar to each other, showing a rather similar status of the microbial community in the different redox states measured. On the other hand, the negative trend of the peak emissions of CH_4 appears very regular and decreases linearly with the degree of anaerobicity reached by soils during incubation (Fig. 4).

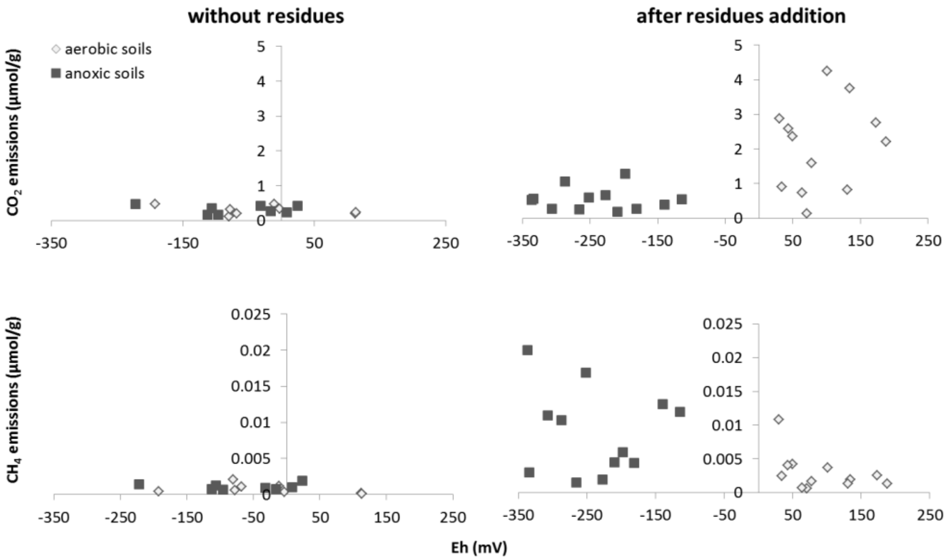

Figure 4. *Comparison between emissions and Eh with and without addition of Limonium residues to soils.*

Conclusions

The alternating flooding conditions, that characterize saltmarsh soils, strongly influence decomposition processes. Anoxic and tidal regimes showed a similar behaviour regarding CO_2 emissions. On the contrary, CH_4 emissions were less different among the three experimental treatments, showing only slightly larger values under completely submerged conditions. Texture appeared an important factor, determining quicker rates in reaching peak values for CO_2 in coarse textured soils and higher CH_4 emissions in fine textured soils. Moreover, soil organic matter content strongly influenced results: larger values refer to higher emissions of both CO_2 and CH_4. The redox status attained by the different soils at the end of the incubation influenced CO_2 release only at positive Eh values, whereas peak emissions of CH_4 decreased steadily when Eh became progressively more positive.

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